

REMARKS

It is respectfully submitted that the amendments submitted herewith function to add new claims 50-54 and to insert the sequence listing and appropriate sequence identifiers into the text of the present application to comply with 37 C.F.R. §1.821 to 1.825. These amendments are made without prejudice, without admission, without surrender of subject matter, and without any intention of creating any estoppel as to equivalents.

It is submitted that new claims 50-54 are in full compliance with the requirements of 35 U.S.C. §112. The amendments and remarks herein are not made for the purpose of patentability within the meaning of 35 U.S.C. §§ 101, 102, 103 or 112; but rather the amendments and remarks are made simply for clarification and to round out the scope of protection to which Applicants are entitled.

Support for the new claims is found throughout the specification, and may specifically be found in example 8 on pages 45-53.

It is respectfully asserted that the sequence disclosure contained in the application now fully complies with the requirements set forth in 37 C.F.R. § 1.821 to § 1.825.

It is respectfully submitted that the Sequence Listing conforms to the requirements of 37 C.F.R. §1.823(b). The Statements required by 37 C.F.R. §1.821(f) and (g) are set forth below.

Pursuant to 37 C.F.R. §1.821 (g), the undersigned hereby states that this submission, filed in accordance with 37 C.F.R. §1.821 (g), does not contain new matter.

Pursuant to 37 C.F.R. §1.821 (f), the undersigned hereby states that the content of the paper and computer readable copies of the Sequence Listing submitted in accordance with 37 C.F.R. §1.821 (c) and (e), respectively, are the same.

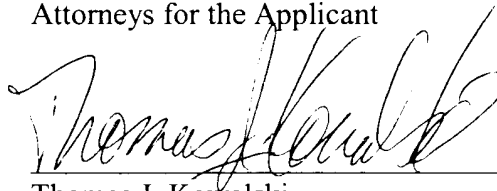
CONCLUSION

In view of the amendments, remarks and enclosures herein, it is respectfully submitted that the application now complies with all requirements set forth in the Notice. Accordingly, reconsideration and withdrawal of the Notice to Comply with Requirements for Patent Applications Containing Nucleotide Sequence and/or Amino Acid Sequence Disclosures ("Notice to Comply") is respectfully requested.

Respectfully submitted,

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By:



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Enclosures: Paper and Diskette copies of Sequence Listing
Copy of Notice to Response To Notice To Comply
Return receipt postcard

The “strength” or “weakness” of doughs are [is] an important aspect of making farinaceous finished products from doughs, including baking. The “strength” or “weakness” of a dough is primarily determined by its content of protein and in particular the content and quality of the gluten protein is an important factor in that respect. Flours with a low protein content are [is] generally characterized as “weak.” Thus, the cohesive, extensible, rubbery mass which is formed by mixing water and weak flour will usually be highly extensible when subjected to stress, but it will not return to its original dimensions when the stress is removed.

Table 3.1. Alignment of lipase 3 peptides with known lipase sequences

LIP_RHIDL (SEQ ID NO: 10)	MVSFISISQGVSLCLLVSSMMLGSSAVPVSGKSGSSNTAVSASDNaALPP	50
LIP_RHIMI (SEQ ID NO: 11)	MVLKQRANYLGFLIVFFTAFLV--EAVPIKRQSNSTVDS-----LLP	40
MDLA_PENCA (SEQ ID NO: 12)	MRLS-----FFTAL-----SAVASLGYALPG	21
N-Terminal	SVSTSTLDELQLFAQWSAAAYXSNN (SEQ ID NO: 20)	
LIP_RHIDL	LISSRCAPPSNKGSKSDLQAEPYNMQKNTIEWYESHGGNLTSGIKRDDNLV	100
LIP_RHIMI	LIPSRTSAPSSSPSTTDPEAPAM-----SRNGPLPS---DVETK	76
MDLA_PENCA	KLQSR-----DVSTSELDQFEFWVQYAAASY-----	47
	..** * 	
LIP_RHIDL	GGMTLDLPSDAPPISLSSSTNSASDGGKVVAATTAQIQEFTKYAGIAATA	150
LIP_RHIMI	YGMALNATSYPDsv-----VQAMSIDGGIRAATSQEINELTYTTLSANS	121
MDLA_PENCA	-----YeadyTAQVGDKL	60
LIP_RHIDL	YCRSVVPGNKWDCVQCQKWVPDgKIITFTT-SLLSDTNGYVLRSDKQKTI	199
LIP_RHIMI	YCRTVIPGATWDCIHCDa-TEDLKIiKtWS-TLIYDTNAMVARGDSEKTI	169
MDLA_PENCA	SCSKG------NCPEVEa--TGATVSYDFSDSTITDTAGYIAVDHTNSAV	102

VHTGFWK (SEQ ID NO: 2)

AWESAADELTSK (SEQ ID NO: 19)

LIP_RHIDL	YLVFRGTNSFRSAITDIVNFSDYKPVKGAKVHAGFLSSYEQVVNDYFPV	249
LIP_RHIMI	YIVFRGSSSIRNW IADLTFVPVSYPPVSGTKVHKGFLDSYGEVQNELVAT	219
MDLA_PENCA	VLAFRGSYSVRNWVADATFVHTNPGLCDGCLAELGFWSSWKLVRDDIIKE	152

	*** * * * * *	
Peptide 2	IK	
LIP_RHIDL	VQEQLTAHPTYKVIVTGHSLGGAQALLAGMDLYQREPRLSPKNLSIFTVG	299
LIP_RHIMI	VLDQFKQYPSYKVAVTGHSLGGATALLCADDLYQREEGLSSSNLFLYTQG	269
MDLA_PENCA	LKEVVAQNPNYELVVVGHSLGAAVATLAATDL -- RGKGYPsakLYAYA--	198
	* * * * * *	
LIP_RHIDL	GPRVGNPTFAYYVESTGPFQRTVHKRDIVPHVPPQSFGFLHPGESWIK	349
LIP_RHIMI	QPRVGDPAFANYVVSTGIPYRRTVNERDIVPHLPAAAFGLHAGEEYWIT	319
MDLA-PENCA	SPRVGNAALAKYITAQGNF-RFHTNDPVPKLPLLSMGYVHVSPEYWIT	247
	*** * * * * *	
LIP_RHIDL	SGTSN - V - - - - QICTSEIETKDCSNSIVPFTSILD - HLSYF - DINEGSC	391
LIP_RHIMI	DNSPETV - - - - QVCTSDLETSDCSNSIVPFTSVLD - HLSYF - GINTGLC	362
MDLA_PENCA	SPNNATVSTSDIKVIDGDVSFDGNTGTGLPLLTDFAHIWYFVQDAGKG	297
LIP_RHIDL	----- L	392
LIP_RHIMI	----- T	363
MDLA_PENCA	PGLPFKRV	305

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Degenerated primers for PCR amplification of a fragment of the lipase gene were designed based on the amino acid sequence of the isolated peptides. The following three PCR primers were synthesized:

C035: TTC CAR YTN TTY GCN CAR TGG (SEQ ID NO: 5)

18 mer 256 mixture, based on the N-terminal sequence QLFAQW. (SEQ ID NO: 21)

C037: GCV GCH SWY TCC CAV GC (SEQ ID NO: 6)

17 mer 216 mixture, based on internal peptide 2 sequence AWESAA (reversed).(SEQ ID NO: 22)

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Table 5.1. (SEQ ID NO: 13)PCR-generated putative *lipA* sequence

(The four amino acid fragments of table 5.1 are contained in SEQ ID NOS: 14-17)

10	20	30	40	50	60
taccgpgntccatt <u>CAGTTGTT</u> CGCGCAATGGTCTGCCGCAGCTTATTGCTCGAATA					
<u>Q L F A Q W S A A A Y C S N</u>					
70	80	90	100	110	120
ATATCGACTCGAAAGAVTCCAACCTTGACATGCACGGCCAACGCCTGTCCATCAGTCGAGG					
<u>N</u> I D S K X S N L T C T A N A C P S V E					
130	140	150	160	170	180
AGGCCAGTACCACGATGCTGCTGCTGGTGGAGTTCGACCTGTATGTCACCTCAGATCGCAGACATAG					
E A S T T M L L E F D L Y V T Q I A D I					
190	200	210	220	230	240
AGCACAGCTAATTGAACAGGACGAACGACTTTTGGAGGCACAGCCGGTTTCCTGGCCGCG					
E H S - L N R T N D F W R H S R F P G R					
250	260	270	280	290	300
G Q H Q Q A A R G R L P G K Q H D - E L					
310	320	330			
ATTGCTAATCYTGACTTCATCCTGGRAGATAACG					
D C - X - L H P X R - (SEQ ID NO: 13)					

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The gene was sequenced using cycle sequencing and conventional sequencing technology. The complete sequence (SEQ ID NO: [8] 18) is shown below in Table 6.1. The sequence has been determined for both strands for the complete coding region and about 100bp upstream and downstream of the coding region. The sequences downstream to the coding region have only been determined on one strand and [contains] contain a few uncertainties. In the sequence as shown below, the intron sequences are indicated as lowercase letters and the N-terminal and the two internal peptides (peptide 1 and peptide 2) are underlined:

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Table 6.1. (SEQ ID NO: 18) The DNA sequence for the lipA gene and flanking sequences

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2    CCNDTTAATCCCCACCGGGGTTCCCGCTCCCGGATGGAGATGGGGCCAAACTGGCAAC
61   CCCCAGTTGCGCAACGGAACAACCGCCGACCCGGAACAAAGGATGCGGATGAGGAGATAC
121  GGTGCCTGATTGCATGGCTGGCTTCATCTGCTATCGTGACAGTGCTCTTTGGGTGAATAT
181  TGTGTCTGACTTACCCCGCTTCTTGCTTTTTTCCCCCTGAGGCCCTGATGGGGAATCGC
241  GGTGGGTAATATGATATGGGTATAAAAGGGAGATCGGAGGTGCAGTTGGATTGAGGCAGT
301  GTGTGTGTGTGCATTGCAGAAGCCCGTTGGTTCGCAAGGTTTTGGTGCCTCGATTGTTTG
361  TATACCGCAAGATGTTCTCTGGACGGTTTGGAGTGCTTTTGACAGCGCTTGCTGCGCTGG
      M F S G R F G V L L T A L A A L
421  GTGCTGCCGCGCCGGCACCGCTTGCTGTGCGGAgtaggtgtgcccgatgtgagatggttg
      G A A A P A P L A V R
481  gatagcactgatgaaggtgaatagTGTCTCGACTTCCACGTTGGATGAGTTGCAATTG
      S V S T S T L D E L Q L
541  TTCGCGCAATGGTCTGCCGACGCTTATTGCTCGAATAATATCGACTCGAAAGACTCCAAC
      F A Q W S A A A Y C S N N I D S K D S N
601  TTGACATGCACGGCCAACGCCTGTCCATCAGTCGAGGAGGCCAGTACCACGATGCTGCTG
      GAGTTTCGACCTgtatgtcactcagatcgagacatagagcacagctaatttgaacagGAC
      E F D L
722  GAACGACTTTGGAGGCACAGCCGTTTCTGCGCGGACAACACCAACAAGCGGCTCGT
      N D F G G T A G F L A A D N T N K R L V
782  GGTGCGCTTCCGGGGAAGCAGCAGCATTGAGAAGTGGATTGCTAATCTTGACTTCATCCT
      V A F R G S S T I E N W I A N L D F I L
842  GGAAGATAACGACGACCTCTGCACCGGCTGCAAGGTCCATACTGGTTTCTGGAAGGCATG
      E D N D D L C T G C K V H T G F W K A W
902  GGAGTCCGCTGCCGACGAAGTACGAGCAAGATCAAGTCTGCGATGAGCACGTATTCCGGG
      E S A A D E L T S K I K S A M S T Y S G
962  CTATACCCTATACTTCAACGGGCACAGTTTGGGCGGCGCATTGGCTACGCTGGGAGCGAC
      Y T L Y F T G H S L G G A L A T L G A T
1022 AGTTCTGCGAAATGACGGATATAGCGTTGAGCTGGTGAGTCCTTCACAAAGGTGATGGAG
      V L R N D G Y S V E L
1082 CGACAATCGGGAACAGACAGTCAATAGTACACCTATGGATGTCCTCGAATCGGAAACTAT
      Y T Y G C P R I G N Y
1142 GCGCTGGCTGAGCATATCACCAGTCAGGGATCTGGGGCCAACTCCGTTTACACACTTG
      A L A E H I T S Q G S G A N F R V T H L
1202 AACGACATCGTCCCGGGGTGCCACCCATGGAAGTTGGATTGAGTCAGCCAAGTCCGGAA
      N D I V P R V P P M D F G F S Q P S P E
1262 TACTGGATCACCAGTGGCAATGGAGCCAGTGTCACGGCGTCGGATATCGAAGTCATCGAG
      Y W I T S G N G A S V T A S D I E V I E
1322 GGAATCAATTCAACGGCGGGAAATGCAGGCGAAGCAACGGTGAGCGTTGTGGCTCACTTG
      G I N S T A G N A G E A T V S V V A H L
1382 TGGTACTTTTTTTCGATTTCAGAGTGCCTGCTATAACTAGACCGACTGTCAGATTAGTGG
      W Y F F A I S E C L L
1442 ACGGGAGAAGGTGACATAAGTAATTAGTATATAATCAGAGCAACCCAGTGGTGGTGATGG
1502 TGGTGAAAGAAGAAACACATTGAGTTCCCATACGKAGCAGWTAAAGCACKTKKGGAGGC
1562 GCTGGTTTCTCCACTTGGCAGTTGGCGGCCATCAATCATCTTCTCTCCTTACTTTTCGT
1622 CCACCACAACCTCCCATCTGCCAGCTGTCGCATCCCCGGGTGCAACAACCTATCGCCTCC
1682 GGGGCTCCGTGGTTCTCCTATATTATCCATCCGACGGCCGACGTTTCACCCTCAACCT
1742 GCGCCGCGGCAAAATCTCCCCGAGTCGGTCAACTCCCTCGAACC GCCCGCCGATCGACC
1802 TCACGACCCCGACCGTCTGYGATYGTCCAACCG

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Table 6.2 Alignment of the lipase 3 sequence with known fungal lipases

LIPASE 3	MFSG ----- RFGVLL ----- TALAA	-15
MDLA_PENCA	MRLS ----- FETAL ----- SAVAS	-14
LIP_RHIDL	MVSFISISQGVSLCLLVSSMMLGSSAVPVSGKSGSSNTAVSADNAALPP	-50
LIP_RHIMI	MVLKQRANYLGFLIVFFTAFLV -- EAVPIKRQSNSTVDS----- LPP	-40
LIPASE 3	L -----	-16
MDLA_PENCA	L -----	-15
LIP_RHIDL	LISSRCAPPSNKGSKSDLQAEPYNMQNTEWYESHGGNLTSGIKRDDNLV	-100
LIP_RHIMI	LIPSRTSAPSSSPSTTDPEAPAM ----- SRNGPLPS ---- DVETK	-76
LIPASE 3	-----GAAAPAPLA-----VRSVSTSTLDELQLFAQWSAAA	-47
MDLA_PENCA	-----GYALPGKLQ-----SRDVSTSELDQFEFVWQYAAAS	-46
LIP_RHIDL	GGMTLDLPSDAPPISLSSSTNSASDGGKVVAATTAQIQEFTKYAGIAATA	-150
LIP_RHIMI	YGMALNATSYPSDV-----VQAMSIDGGIRAATSQEINELTYTTLSANS	-121
LIPASE 3	YCSNNIDSK-DSNLTCTANACPSVEEASTTMLLEFDLTNDFGGTAGFLAA	-96
MDLA_PENCA	YYEADYTAQVGDKLSCSKGNCPEVEATGATVSYDFS-DSTITDTAGYIAV	-95
LIP_RHIDL	YCRSVVP---GNKWDCVQ---CQKWVPDGKIIT---TFTSLLSDTNGYVLR	-192
LIP_RHIMI	YCRTVIP---GATWDCIH---CDA-TEDLKIHK---TWSTLIYDTNAMVAR	-162
LIPASE 3	DNTNKRLVVAFRGSSSTIENWIANLDFILEDNDDLCTGCKVHTGFWKAWES	-146
MDLA_PENCA	DHTNSAVVLAFRGSYSVRNWVADATFV-HTNPGLCDGCLAELGFWSSWKL	-144
LIP_RHIDL	SDKQKTIYLVFRGTNSFRSAITDIVFNFSYKPV-KGAKVHAGFLSSYEQ	-241
LIP_RHIMI	GDSEKTIYIVFRGSSSIRNWIADLTFVPVSYPPV-SGTVHKGFLLDSYGE	-211
LIPASE 3	AADELTSKIKSAMSTYSGYTLYFTGHSLGGALATLGATVL---RNDGY-SV	-193
MDLA_PENCA	VRDDIIKELKEVVAQNPNYELVVVGHSLGAAVATLAATDL---RGKGYPSA	-192
LIP_RHIDL	VVNDYFPVVQEQLTAHPTYKVIVTGHSLGGAQALLAGMDLYQREPRLSPK	-291
LIP_RHIMI	VQNELVATVLDQFKQYPSYKVAVTGHSLGGATALLCALDLYQREEGLSSS	-261
LIPASE 3	ELYTY---GCPRIGNYALAEHITSQGSGANFRVTHLNDIVPRVPPMDFGFS	-241
MDLA_PENCA	KLYAY -- ASPRVGNAALAKYITAQGN -- NFRFHTNDPVPKPLLLSMGYV	-238
LIP_RHIDL	NLSIFTVGGPRVGNPTFAYYVESTGIPFQ-RTVHKRDIVPHVPPQSFGFL	-340
LIP_RHIMI	NFLYTQGGQPRVGDPAFANYVVSTGIPYR-RTVNERDIPVPHLPPAAFGL	-310
LIPASE 3	QPSPEYWITSGNGASVTASDIEVIEGINSTAGNAGEATVSVV---AHLWY	-288
MDLA_PENCA	HVSPEYWITSPNNATVSTSDIKVIDGDVSFDGNTGTGLPLLTDFEAHIWY	-288
LIP_RHIDL	HPGVESWIKSGTSN-VQICTSEIE-----TKDCSNSIVPETSILDHLSY	-383
LIP_RHIMI	HAGEEYWITDNSPETVQVCTSDLE-----TSDCSNSIVPFTSVLDHLSY	-354

LIPASE 3	FFAISECL- - - - -L	-297 (SEQ ID NO: 9)
MDLA_PENCA	FVQVDAGKGPGLPFKRV	-305 (SEQ ID NO: 12)
LIP_RHIDL	F - DINEGSC- - - - -L	-392 (SEQ ID NO: 10)
LIP_RHIMI	F - GINTGLO- - - - -T	-363 (SEQ ID NO: 11)

* ...

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Table 6.3 Alignment of coding sequence of the *lipA* gene and gene coding for mono-diacyl lipase from *Penicillium camemberti*

LIPASE 3	MFSGRFGVLLTALAALGAAAPAPLAVRSVSTSTLDELQLFAQWSAAAYCS	-50
MDLA_PENCA	MRLSFFTAL - SAVASLGYALPGKLQSRDVSTSELDQFEFWVQYAAASYYE	-49
LIPASE 3	NN I DSK - DSNLTCTANACPSVEEASTTMLLEFDLTNDFGGTAGFLAADNT	-99
MDLA_PENCA	ADYTAQVGDKLSCSKGNCPEVEATGATVSYDFS - DSTITDTAGY IAVDHT	-98
LIPASE 3	NKRLVVAFRGSSTIENW I ANLDFILEDNDDLCTGCKVHTGFWKAWESAAD	-149
MDLA_PENCA	NSAVVLAFRGSYSVRNWWADATFV-HTNPGLCDGCLAELGFWSSWKLVRD	-147
LIPASE 3	ELTSKIKSAMSTYSGYTLYFTGHSLGGALATLGATVLRNDGY-SVELYTY	-198
MDLA_PENCA	DIIKELKEVVAQNPNYELVVVGHSLGAAVATLAATDLRGKGYPYSAKLYAY	-197
LIPASE 3	GCPR IGNYALAEHITSQGSGANFRVTHLNDIVPRVPPMDFGFSQPSPEYW	-248
MDLA_PENCA	ASPRVGNAALAKYITAQGN- - NFRFHTNDPVPKPLLLSMGYVHVSPEYW	-245
LIPASE 3	ITSGNGASVTASDIEVIEGINSTAGNAGEATVSVV - - - AHLWYFFAISEC	-295
MDLA_PENCA	ITSPNNATVSTSDIKVIDGDVSFDGNTGTGLPLLTDTFEAHIWYFVQVDAG	-295
LIPASE 3	L-----L -297 (SEQ ID NO: 9)	
MDLA_PENCA	KGPGLPFKRV -305 (SEQ ID NO: 12)	

Identity: 126 amino acids (42.42%)

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Table 6.4: Amino acid sequence of the precursor of lipase 3 (SEQ ID NO: 9)

	5	10	15	20	25	30
1	M	F	S	G	R	F
	G	V	L	L	T	A
	A	A	L	G	A	A
	P	A	P	A	P	L
	A	V	R	S	V	S
31	T	S	T	L	D	E
	L	Q	L	F	A	Q
	W	S	A	A	A	Y
	C	S	N	N	I	D
	S	K	D	S	N	L
61	T	C	T	A	N	A
	C	P	S	V	E	E
	A	S	T	T	M	L
	L	E	F	D	L	T
	N	D	F	G	G	T
91	A	G	F	L	A	A
	D	N	T	N	K	R
	L	V	V	A	F	R
	G	S	S	T	I	E
	N	W	I	A	N	L
121	D	F	I	E	D	N
	D	D	L	C	T	G
	C	K	V	H	T	G
	F	W	K	A	W	E
	S	A	A	D	E	
151	L	T	S	K	I	K
	S	A	M	S	T	Y
	S	G	Y	T	L	Y
	F	T	G	H	S	L
	G	G	A	L	A	T
181	L	G	A	T	V	L
	R	N	D	G	Y	S
	V	E	L	Y	T	Y
	G	C	P	R	I	G
	N	Y	A	L	A	E
211	H	I	T	S	Q	G
	S	G	A	N	F	R
	V	T	H	L	N	D
	I	V	P	R	V	P
	P	M	D	F	G	F
241	S	Q	P	S	P	E
	Y	W	I	T	S	G
	N	G	A	S	V	T
	A	S	D	I	E	V
	I	E	G	I	N	S
271	T	A	G	N	A	G
	E	A	T	V	S	V
	V	A	H	L	W	Y
	F	F	A	I	S	E
	C	L	L			

Number of residues: 297